

Factors Affecting Phytochemical Content In Some Sweet Sorghum Varieties

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ABSTRACT

This study aimed to identify the bioactive compounds existing in sweet sorghum products (juice and syrup) that help increase their usage in the food industry. There are a lot of health benefits that can be derived from sweet sorghum that can be used in developing the food products that can be benefitted by target consumers. To achieve this purpose, it is important to find out factors that affect the quantity and kind of bioactive compounds, mainly (phenolic acids and flavonoids), among different sweet sorghum products (juice and syrup). Hence, a field trial was conducted in 2022 in Agricultural Research Center, Ministry of Agriculture and Reclaimed Land, Giza Governorate, Egypt, to assess the influence of two harvest stages, i.e., milk and dough stages, and juice concentration treatments, i.e., 73, 75, 78, and 80° Brix (measured by hand refractometer), on the bioactive compounds of three sweet sorghum (*Sorghum bicolor*, L., Moench) varieties (Rex, Roma, and Welly). Results revealed that at the milk harvesting stage, there was a reverse relationship between sucrose, glucose, and fructose juice content, while during the dough stage, sugar accumulation increased, especially for sucrose, which was accompanied by fructose and glucose as well. Generally, phenolic compounds increased at the dough stage in the three studied varieties, while the flavonoid content of the juice increased during the dough stage in both Welly and Roma varieties. The concentration process of the juice led to a noticeable increase in phenolic content, especially for the Rex variety, which exhibited superior behavior among the other varieties.

1. Introduction

Sweet sorghum (*Sorghum bicolor* L. Moench) is an individual plant material which simultaneously produces grains with high content of starch, sugary juice and content of biomass lignocellulose. Its pliability in accommodating ramified climatic and soil type participates to its state as a bivotal food (Mbulwe and Ajayi, 2020). On the world, Sweet sorghum is the fifth important cereal crop, versatility of sorghum makes it remarkable components in agriculture economy (Habyarimana et al., 2020). Sorghum demands lower requirements of agriculture compared to other crops, making it an appropriate choice for farmers. Sorghum is considered an optimal crop in face of climate change. Sweet sorghum has high nutritional value contribute to health benefits where, it contains protein, carbohydrate and bioactive compo-

nents (vitamins and polyphenols). Yun-long et al. (2006) revealed that dough stage is suitable to measure sugars content of sweet sorghum stalk. Sweet sorghum varieties have a diversity of phytochemicals such as phenolic acids and flavonoids which include tannins and anthocyanins with a potential to significantly impact human health in, phytochemical in sweet sorghum have high antioxidant activity against different free radicals (Dicko et al., 2005). Polyphenols in sorghum are phenolic acid (benzoic, cinnamic, their derivatives) and flavonoids (tannins and anthocyanins), (Krueger et al., 2003). Previous studies recommended that the evaluation of sorghum cultivars based on bioactive constitution is necessary for selecting cultivars during syrup preparation.

However, further studies on phytochemical and biochemical changes during juice processing into syrup may enhance the importance of these parameters in considering suitable cultivars for syrup production. This can also contribute to increase the market value of sweet sorghum juice and syrup as consumable sources. Among the five sorghum juices analyzed, all (sugars, antioxidants, and phenols) were found to be high in the E10 variety. Kurella et al. (2020) stated that sweet sorghum cultivars like E10, which exhibit high phytochemical and carbohydrate content, can be utilized as a raw material for high-quality syrup. It is crucial to consider the phytochemical and biochemical characteristics of sorghum cultivars when determining the optimal variety for syrup production, as this can influence health benefits and promote their wider use as a food source. Also, sweet sorghum juice can be an excellent source of phenols, antioxidant phytochemicals, and it may be beneficial to select suitable varieties for syrup production based on phytochemical and biochemical indicators. They also concluded that the E10 sweet sorghum variety surpassed the other four varieties in sugars, protein, antioxidants, and phenol content. Using Vacuum Falling Film Evaporator (FFE) at temperatures 70, 80, and 90°C as a single treatment or combined with rotary Evaporator (RE) at 60, 70, and 80°C to produce sweet sorghum syrup. The highest brix value was recorded by single FFE (90°C) and combined FFE(90°C)-RE(80°C), respectively (44.2° Brix and 87.53°Brix) where TSS and reducing sugar of sorghum syrup resulted from combined FFE(90°C)-RE(80°C) increased by five times and 2.4 times, respectively as compared to raw juice indicating that the sucrose of sorghum syrup was stable during evaporation (Yuwono et al., 2020). Egglestone et al. (2021) found that samples of sweet sorghum syrup contained phenolic compounds (phenolic acids and flavonoids), the prevalent components were phenolic acids which included Ellagic (335–1177 $\mu\text{g g}^{-1}$), protocatechuic (53–485 $\mu\text{g g}^{-1}$), Sinapic (21– 3654 $\mu\text{g g}^{-1}$), vanillic (10– 96 $\mu\text{g g}^{-1}$) and low concentrations of chlorogenic and Gallic acid. While flavonoids such as

Catechin (122-2670 $\mu\text{g g}^{-1}$), Naringin and Apigenin with low concentrations. Massey (2014) discovered that the sweet sorghum stalk, particularly the dermal layer, contained significant amounts of bioactive phytochemicals, including phenolic acids, anthocyanidins, and flavones, which demonstrated in vitro and in vivo anticancer activities against colon cancer HCT116 cells and colon cancer stem cells (CCSCs). Uchimiya and Knoll (2020) concluded that the No.5 Gambela cultivar was enriched with polyphenols and exhibited a higher Epa (potential a) compared to the N109A×Chinese cultivar, which is an amino acid-enriched cultivar. They also reported that juice extracted from sweet sorghum contains a complex mixture of polyphenols, with quercetin particularly abundant in the No.5 Gambela cultivar. This cultivar could provide an antioxidant-rich juice suitable for conversion into edible syrup.

El-Geddawy (2019) found that phenolic compounds varied among different sweet sorghum varieties. The juice of the Welly variety had the highest content of pyrogallol and 3-OH-tyrosol (89.06 and 20.97 ppm, respectively). The juice of the Rex variety had the highest content of gallic acid (10.83 ppm). Additionally, the juice of the Rex variety exhibited high levels of pyrogallol, chlorogenic acid, and 3-OH-tyrosol (53.71, 23.06, and 17.22 ppm, respectively). The highest content of phenolic compounds, especially chlorogenic acid, p-OH-benzoic acid, and e-vanillic acid, was found in Roma sweet sorghum juice (42.53, 28.89, and 51.41 ppm, respectively). In addition to sweet sorghum Roma juice had the highest content of flavonoid compounds, followed by Rex juice, particularly naringin (12.92 and 5.63 ppm), rutin (10.85 and 4.34 ppm), quercitrin (8.37 and 3.36 ppm), rosmarinic acid (3.32 and 2.41 ppm), and hesperidin (0.77 and 0.31 ppm), respectively. Meanwhile, hesperidin, naringin, rutin, and quercitrin were the dominant flavonoids in Welly sweet sorghum juice (8.26, 4.55, 3.57, and 3.17 ppm, respectively). Regarding concentration effects, the Rex variety had the highest content of almost all phenolic compounds. The flavonoid content of different syrup samples

increased after concentration, with the Rex variety having the highest content of all flavonoids compared to other sweet sorghum varieties. The dominant flavonoids were naringin, hesperidin, hesperetin, rosmarinic acid, and quercitrin (110.6, 47.35, 46.31, 37.30, and 21.36 ppm, respectively). Sucrose is the predominant sugar in the syrups of sweet sorghum Rex variety. After the concentration process, sugar fractions in the syrups, especially glucose and sucrose, increased, with sucrose remaining the predominant sugar in the syrups of sweet sorghum Rex variety, followed by glucose (El-Geddawy, 2019). Almodares et al. (2007) studied the effect of maturity stage and variety on sucrose percentage. The results showed that the Rio sweet sorghum variety had the highest sucrose content, followed by Vespa variety, while the lowest values were recorded by IS2325 variety (11.53%, 7.38%, and 6.57%, respectively). It was noted that the highest sucrose content was found at the physiological maturity stage (10.80%), while the lowest content was observed at the flowering stage (6.07%). Sweet sorghum stalks were harvested at the dough stage, and the results showed that the major disaccharide in sweet sorghum juice was sucrose, ranging between 2.58% and 5.48%. The glucose content varied between 1.12% and 2.94%, fructose ranged between 1.05% and 2.39%, as reported by Kumar et al. (2013). Thus the present investigation attempts to identify the phytochemical compounds (phenolic acid and flavonoids) in sweet sorghum juice and syrup during two maturity stages (milk and dough stage) and four brix degree for concentration using three different sweet sorghum cultivars. So, suitable harvesting stage and/or cultivars for juice and syrup rich in bioactive constitution can be selected and used in developing the food products that can benefit target consumers.

2. Materials and Methods

The field trial was carried out at the Agricultural Research Center, Giza, Governorate, Egypt, in May during 2022 season. The study aimed to identify the phytochemical compounds in sweet sorghum juice and syrup by studying the influence of two harvest

stages (milk and dough) and four juice concentration treatments (73, 75, 78, and 80 Brix, measured by hand refractometer) on these compounds for three sweet sorghum (*Sorghum bicolor*, L., Moench) varieties (Rex, Roma, and Welly).

Juice extraction and Syrup preparation

The millable stalks of the three sweet sorghum varieties were stripped, and squeezed using an electric horizontal three-roller mill with high extraction efficiency. Subsequently, suspended matter was removed through two filtration steps using wire screen and fine mesh screen to get clear juice (El-Geddawy, 2019). The cleared juice was evaporated in stainless steel pan, as increasing the temperature some substances (non sugars and proteins) were coagulated, then skimming was started to remove them, after removing scum, syrup clarity was observed. Brix was measured by "Hand Refractometer", after heating was stopped, the syrup was cooled and poured into sterilized bottles.

Quantitative determination of sugars using HPLC

Sugars content in samples (juice and syrup) were estimated according to Dolciotti et al. (1998) and long et al. (2006) using high-performance liquid chromatography (HPLC) Knauer, Germany equipped with two pumps, RI detector, UV detector, column oven and clarity-chrom software as described in following steps: Five gram of sample (juice or syrup) were dissolved in 12 ml methanol HPLC grade, quantitatively transferred to measuring flask 50 ml completed to the mark with HPLC grade water, sonicated for 20 min, Filtered through PTFE filter (0.2 mm), Kept at 0° C until analysis. The flow rate was adjusted at 2 ml/min, the column was Luna NH2 column for sugar identification, The column oven temperature kept constant at 40° C, the RI 58 detector operated at room temperature, the mobile phase was Acetonitrile: HPLC grade (80:20 v/v).

Determination of phenolic and flavonoid compounds

To identify phenolic acids and flavonoids in the extracted juice and its concentrate (syrup) from the

three sweet sorghum varieties, high-performance liquid chromatography (HPLC) analysis was performed. Samples were prepared according to the method described by Jakopič et al. (2009). Chromatograms were analyzed at 280 nm to detect phenolic acids and at 330 nm for flavonoids. Compound identification and quantification were achieved by comparing peak areas to external standards, as outlined by Schieber et al. (2001).

Statistical Analysis

All data were expressed as mean values. Statistical analysis was performed using SPSS (16.0) program, one way analysis of variance or two way analysis of variance (ANOVA) followed by Duncan's Multiple Range Test with $P \leq 0.05$ being considered statistically significant, (Snedecor and Cochran, 1980).

3. Results and Discussion

Effect of different varieties and harvesting stages on sugar fractions of sweet sorghum products (juice and syrup)

Effect of different varieties and harvesting stages on sugar fractions of sweet sorghum products (juice and syrup). Sugars content analysis was conducted using HPLC (Table 1). Results showed that the harvest stage significantly influenced sugars concentration in the juice extracted from the studied sweet sorghum varieties. At the milk stage, Roma variety exhibited the highest sucrose content (7.47

g/100 g), accompanied by the lowest glucose and fructose levels (4.71 and 3.80 g/100 g, respectively). Welly variety displayed lower sucrose but higher glucose and fructose concentrations (1.28, 5.64, and 4.79 g/100 g, respectively). Notably, sucrose was undetectable in Rex variety, which recorded the highest glucose and fructose levels (8.38 and 6.93 g/100g). At the dough stage, all sugar levels increased except for sucrose in Roma variety. Welly variety exhibited the highest sucrose content (13.06 g/100g), followed by Rex (8.37g/100 g), which recorded the highest glucose and fructose concentrations (8.83 and 7.97g/100 g, respectively). The concentration process led to an increase in sugars content in syrup samples, concerning Rex variety showed the most pronounced elevation in sucrose, glucose, and fructose (10.12, 11.00, and 40.85 g/100g, respectively). These variations in sugars concentration can be attributed to factors such as the rate of phloem transport, nutrient storage, environmental conditions, and plant developmental stage. The relationship between sucrose and harvest time appears stronger than that between glucose and fructose. These findings align with Li et al. (2013) who found that *Sorghum bicolor* accumulates sucrose in the stem and recommended *Sorghum bicolor* as a source of syrup production. Also, Willis et al. (2013) who reported high sucrose levels followed by glucose and fructose in sweet sorghum syrup.

Table 1. Fractionation of sugars in juice and syrup of three sweet sorghum varieties in two harvesting stages

Type of sample	Maturity stage	Sugars%	Variety		
			Roma	Rex	Welly
Sweet sorghum juice	Milk stage	Fructose	3.80 ^{cC}	6.93 ^{bA}	4.79 ^{bB}
		Glucose	4.71 ^{bC}	8.38 ^{aA}	5.64 ^{aB}
		Sucrose	7.47 ^{aA}	ND	1.28 ^{cB}
	Dough stage	Fructose	6.62 ^{aB}	7.97 ^{cA}	6.45 ^{bC}
		Glucose	6.22 ^{bC}	8.83 ^{aA}	6.42 ^{cB}
		Sucrose	6.62 ^{aC}	8.37 ^{bB}	13.06 ^{aA}
Sweet sorghum syrup (Rex variety)	Dough stage	Fructose	6.39 ^c	10.12 ^c	6.46 ^c
		Glucose	8.36 ^b	11.00 ^b	7.00 ^b
		Sucrose	38.73 ^a	40.85 ^a	28.07 ^a

ND: Not Detected - Means followed by different small letters in the same column (effect of variety) are significantly by Duncan's multiple test ($P \leq 0.05$).

- Means followed by different capital letters in the same row of same parameter (effect of harvesting stage) are significantly by Duncan's multiple test ($P \leq 0.05$)

Effect of different varieties and harvesting stages on phenolic and flavonoid Content of sweet sorghum juice

Table 2 and 3 revealed that phenolic and flavonoid compounds significantly affected by varieties and harvesting stage. Syringic acid, known for its therapeutic properties against various diseases including neurodegenerative disorders, liver damage, and diabetes, as well as its antimicrobial and anti-inflammatory effects, exhibited high concentrations during the milk stage in all three varieties (161.24, 101.52, and 36.88 ppm for Rex, Welly, and Roma, respectively). However, a significant decrease was observed in syringic acid content during the dough stage (1.48, 2.55, and 4.51 ppm for Rex, Welly, and Roma, respectively). Similar trends were noted for protocatechuic acid, which was abundant in Rex variety (53.16 ppm), and pyrogallol, with higher levels during the milk stage (60.79, and 35.70 ppm for Roma and Welly, respectively). Overall, phenolic compound levels increased in all three varieties during the dough stage. These findings align with Chen et al. (2022) who extracted some phenolic compounds from sweet sorghum fresh stalks that showed strong inhibiting ability against food-borne pathogens i.e. (gallic, gentisic, trans-ferulic, caffeic, coumaric, chlorogenic, and phydroxybenzoic acids). Flavonoid content exhibited varied responses to harvest stage. While Welly and Roma varieties showed increased flavonoid levels during the dough stage, Rex variety displayed inconsistent changes with some flavonoids increasing and others decreasing. Hesperidin was the predominant flavonoid among all varieties during the milk stage, followed by naringin (16.18, 13.61, and 6.76 ppm) and (8.33, 6.35 and 3.27 ppm for Rex, Roma and Welly, respectively). Likewise, during the dough stage, epicatechin was the predominant flavonoid in all varieties, followed by hesperidin, rutin and naringin (13.95, 10.05 and 9.47 ppm for Welly; 6.33, 5.46 and 5.33ppm for Rex, respectively), then naringin, catechol, rutin, quercetin, and hesperidin (12.44, 10.50, 7.72, 7.26, and 7.16ppm, respectively). The observed variations in flavonoid content among sweet sorghum varieties align with previous

research by Herrmann (1976) and Kurella et al. (2020), which attributed these differences to genetic factors, environmental conditions, maturity stage, cultivar, fertilization rates, and plant part, among other factors influencing the quantitative variation of bioactive compounds.

Effect of Concentration on phenolic and flavonoid Content

Data presented in Table 4 demonstrated that concentration process at total soluble solids (73%) significantly increased phenolic content. Pyrogallol emerged as the predominant phenolic compound in all three varieties (1530.87, 1022.87, and 353.03 ppm), followed by e-vanillic acid (250.56, 522.92 and 193.52 ppm) for Welly, Rex, and Roma, respectively. then, caffeine (21.41 ppm) in welly, p-OH-benzoic acid (305.22 ppm for Rex) and chlorogenic (137.11 ppm) in Roma. These results are in harmony with those of Payet et al. (2006). A similar trend was observed for flavonoids (Table 5), with concentration enhancing their levels in the juice. Naringin emerged as the predominant flavonoid in syrup of Rex and welly varieties (156.92 and 78.82 ppm), followed by catechol (136.09 and 50.54 ppm), rutin (92.52 in Rex), hesperidin (46.47 ppm in welly). On the other hand, catechol was the predominant flavonoid in syrup of Roma followed by naringin and catechin, respectively (149.3, 93.71 and 49.31, respectively). On the contrary Apegnin flavonoids recorded the lowest values in syrup of Welly and Roma varieties while, 7-OH-flavon the lowest values in syrup of Welly. Generally, sweet sorghum varieties differed in flavonoids content, where Rex sweet sorghum variety demonstrated superior flavonoid content compared to the other two varieties. Overall, phenolic and flavonoid concentrations in sweet sorghum syrup exhibited significant variations among the three varieties. The levels of Phenolic acids ranged from 3.56 to 1530.87ppm, 6.93 to 1022.87ppm, and 4.3 to 353.03ppm, on the other side, flavonoids levels varied from 0.32 to 78.82ppm, 1.17 to 156.95ppm, and 0.20 to 149.30ppm for Welly, Rex, and Roma varieties, respectively.

Table 2. Fractionation of phenolic acids in juice of three sweet sorghum varieties in two harvesting stages

phenolic acid (ppm)	Variety					
	Rex		Roma		Welly	
	Milk	Dough	Milk	Dough	Milk	dough
Gallic	0.51 ^{qB}	1.17 ^{mA}	0.69 ^{pB}	0.85 ^{qA}	0.89 ^{lB}	1.57 ^{pA}
Pyrogallol	36.45 ^{cB}	48.13 ^{aA}	60.79 ^{aA}	43.60 ^{bB}	35.70 ^{bA}	22.13 ^{cB}
4-Amino-benzoic	0.57 ^{pB}	7.58 ^{fA}	0.45 ^{sB}	11.60 ^{fA}	0.31 ^{qB}	12.18 ^{fA}
Protocatchuic	53.16 ^{bA}	1.49 ^{lB}	1.72 ^{lB}	6.68 ^{iA}	9.16 ^{cA}	2.52 ^{lB}
Chlorogenic	7.23 ^{eB}	17.36 ^{cA}	6.04 ^{eB}	38.96 ^{cA}	2.04 ^{gB}	42.45 ^{bA}
Rosmarinic	0.76 ^{oB}	3.36 ^{hA}	2.16 ^{kB}	3.21 ^{kA}	0.37 ^{pB}	6.64 ^{hA}
Caffeine	3.78 ^{iA}	3.32 ^{iB}	5.81 ^{fA}	2.77 ^{mB}	1.46 ^{iB}	2.65 ^{kA}
P-OH- benzoic	1.98 ^{kA}	0.51 ^{sB}	3.44 ^{iA}	ND	1.39 ^{jA}	1.26 ^{rB}
Caffeic	2.79 ^{iB}	10.21 ^{dA}	2.86 ^{jB}	24.37 ^{dA}	2.40 ^{fB}	16.08 ^{cA}
Vanillic	1.59 ^{mA}	0.86 ^{oB}	1.64 ^{mB}	1.71 ^{oA}	1.23 ^{kA}	0.90 ^{tB}
Ferulic	3.14 ^{jA}	1.77 ^{kB}	4.07 ^{hB}	11.45 ^{gA}	1.79 ^{hB}	3.30 ^{jA}
Iso-ferulic	1.75 ^{lB}	3.79 ^{gA}	1.32 ^{nB}	4.55 ^{jA}	0.83 ^{mB}	7.97 ^{gA}
e-vanillic	ND	1.08 ^{nA}	ND	2.42 ^{nA}	ND	2.37 ^{nA}
Reversetrol	ND	23.88 ^{bA}	ND	48.89 ^{aA}	ND	50.61 ^{aA}
Ellagic	1.25 ^{nA}	0.69 ^{rB}	1.30 ^{oB}	1.58 ^{pA}	0.47 ^{oB}	1.52 ^{qA}
α -coumaric	0.17 ^{sB}	0.81 ^{pA}	0.27 ^{tB}	2.99 ^{lA}	0.18 ^{sB}	1.67 ^{oA}
Benzoic	6.63 ^{fA}	0.24 ^{uB}	6.3 ^{dA}	0.15 ^{tB}	6.78 ^{nA}	0.55 ^{wB}
Salicylic	4.92 ^{gB}	9.29 ^{eA}	4.55 ^{gB}	17.64 ^{eA}	0.77 ^{nB}	20.12 ^{dA}
3,4,5- mthoxy-cinnamic	0.53 ^{qB}	3.01 ^{jA}	0.50 ^{rB}	7.38 ^{hA}	0.35 ^{pB}	5.60 ^{iA}
Coumarin	0.40 ^{rA}	0.28 ^{tA}	0.60 ^{qA}	0.59 ^{rA}	0.27 ^{rB}	0.59 ^{vA}
p-coumaric	8.76 ^{dA}	0.02 ^{ovB}	9.61 ^{cA}	0.60 ^{rB}	4.67 ^{eA}	0.69 ^{uB}
Cinnamic	ND	0.29 ^{tA}	0.0016 ^{uB}	0.30 ^{sA}	0.0004 ^{tB}	0.99 ^{sA}
3-OH-Tyrosol	ND	0.77 ^{qA}	ND	1.57 ^{pA}	ND	1.97 ^{nA}
Syringic	161.24 ^{aA}	1.48 ^{lB}	36.88 ^{bA}	4.51 ^{jB}	101.52 ^{aA}	2.55 ^{lB}

- ND: Not Detected - Means followed by different small letters in the same column (effect of variety on Phenolic compound concentration) are significantly by Duncan's multiple test ($P \leq 0.05$).

- Means followed by different capital letters in the same raw of same parameter (effect of harvesting stage) are significantly by Duncan's multiple test ($P \leq 0.05$)

Table 3. Fractionation of flavonoids in juice of three sweet sorghum varieties in two harvesting stages

Variety Flavonoids (ppm)	Rex		Roma		Welly	
	Milk	Dough	milk	Dough	Milk	Dough
	Naringin	8.33 ^{dA}	5.33 ^{dB}	6.35 ^{dB}	12.44 ^{bA}	3.27 ^{cB}
Rutin	2.12 ^{fB}	5.46 ^{cA}	1.53 ^{fB}	7.72 ^{dA}	2.79 ^{dB}	10.05 ^{cA}
Hisperdin	16.18 ^{aA}	6.33 ^{bB}	13.61 ^{aA}	7.17 ^{fB}	6.76 ^{bB}	13.95 ^{bA}
Catechol	9.50 ^{cA}	4.13 ^{cB}	9.07 ^{cB}	10.50 ^{cA}	10.01 ^{aA}	5.11 ^{fB}
Catechein	9.98 ^{bA}	4.13 ^{cB}	10.50 ^{bA}	3.32 ^{gB}	2.04 ^{eB}	5.11 ^{fA}
Epicatechein	4.10 ^{eB}	14.09 ^{aA}	3.23 ^{eB}	30.37 ^{aA}	1.50 ^{fB}	26.05 ^{aA}
Quercetrin	1.05 ^{gB}	3.27 ^{fA}	0.94 ^{gB}	7.26 ^{cA}	0.46 ^{gB}	6.39 ^{eA}
Quercetin	0.29 ^{hA}	0.13 ^{iB}	0.23 ^{hB}	0.42 ^{jA}	0.06 ^{kB}	0.44 ^{iA}
Narengenin	0.25 ^{hA}	0.06 ^{jB}	0.15 ^{jA}	0.15 ^{kA}	0.11 ^{iB}	0.16 ^{jA}
Kampherol	0.07 ^{ijB}	0.35 ^{gA}	0.04 ^{lB}	0.83 ^{hA}	0.02 ^{lB}	0.67 ^{gA}
Hisperdin	0.17 ^{hiA}	0.16 ^{hA}	0.13 ^{kB}	0.59 ^{iA}	0.07 ^{lB}	0.59 ^{hA}
Apegnin	ND	0.02 ^{kA}	0.00 ^{mB}	0.08 ^{lA}	0.15 ^{hA}	0.12 ^{kB}
7-OH flavon	0.02 ^{jA}	0.02 ^{kA}	ND	0.07 ^{lA}	0.02 ^{lB}	0.05 ^{lA}
Luteolin	ND	ND	0.19 ^{iA}	ND	0.19 ^{fA}	ND

- ND: Not Detected

- Means followed by different small letters in the same column (effect of variety on flavonoid compound concentration) are significantly by Duncan's multiple test ($P \leq 0.05$).

- Means followed by different capital letters in the same raw of same parameter (effect of harvesting stage) are significantly by Duncan's multiple test ($P \leq 0.05$)

Table 4. Effect of concentration (TSS% 73) on bioactive compounds(phenolic acid)

Phenolic compound, (ppm)	Variety		
	Welly	Rex	Roma
Gallic	6.65 ^{tC}	26.51 ^{qA}	9.78 ^{rB}
Pyrogallol	1530.87 ^{aA}	1022.87 ^{aB}	353.03 ^{aC}
4-Amino-benzoic	26.59 ^{nB}	28.99 ^{pA}	9.41 ^{sC}
Protocatechuic	80.88 ^{hB}	92.67 ^{gA}	53.42 ^{hC}
Chlorogenic	113.31 ^{gB}	59.62 ^{iC}	137.11 ^{eA}
Rosmarinic	8.31 ^{sA}	6.93 ^{uC}	7.50 ^{tB}
Caffeine	210.41 ^{eA}	128.87 ^{eB}	10.18 ^{pC}
P-OH- benzoic	143.2 ^{eB}	305.22 ^{eA}	89.73 ^{dC}
Caffeic	139.55 ^{fA}	29.08 ^{pC}	54.46 ^{gB}
Vanillic	53.79 ^{jA}	36.92 ^{nB}	33.34 ^{kC}
Ferulic	147.01 ^{dB}	275.89 ^{dA}	52.37 ^{iC}
Iso-ferulic	22.93 ^{pC}	67.64 ^{hA}	26.45 ^{mB}
e-vanillic	250.56 ^{bB}	522.98 ^{bA}	193.52 ^{bC}
Reversetrol	29.60 ^{iC}	50.02 ^{kA}	31.75 ^{lB}
Ellagic	34.33 ^{kC}	105.87 ^{fA}	66.67 ^{fB}
Alpha-coumaric	17.19 ^{qB}	20.38 ^{rA}	10.15 ^{qC}
Benzoic	ND	63.76 ^{iA}	50.67 ^{jB}
Salicylic	57.35 ^{iB}	34.84 ^{oC}	68.75 ^{eA}
3,4,5- mthoxy-cinnamic	25.09 ^{oC}	48.92 ^{lA}	25.59 ^{nB}
Coumarin	11.16 ^{rA}	7.00 ^{tB}	ND
p-coumaric	28.78 ^{mB}	48.66 ^{mA}	11.92 ^{oC}
Cinnamic	3.56 ^{uC}	8.30 ^{sA}	4.30 ^{uB}

- ND: Not Detected

- Means followed by different small letters in the same column (effect of variety on Phenolic compound concentration) are significantly by Duncan's multiple test ($P \leq 0.05$).

- Means followed by different capital letters in the same row of same parameter (effect within variety) are significantly by Duncan's multiple test ($P \leq 0.05$)

Table 5. Effect of concentration (TSS 73%) on bioactive compounds (flavonoids)

Flavonoid content (ppm)	Varieties		
	Welly	Rex	Roma
Naringin	78.82 ^{aC}	156.95 ^{aA}	93.71 ^{bB}
Rutin	21.54 ^{fB}	92.52 ^{eA}	17.13 ^{fC}
Hesperdin	23.52 ^{eB}	44.09 ^{gA}	22.63 ^{eC}
Catechol	50.54 ^{bC}	136.09 ^{bB}	149.3 ^{aA}
Catechein	39.50 ^{dC}	58.69 ^{eA}	49.31 ^{eB}
Quercetrin	13.38 ^{gB}	52.09 ^{fA}	12.48 ^{gC}
Quercetin	2.78 ^{iC}	7.80 ^{iA}	2.82 ^{iB}
Narengenin	2.98 ^{hC}	6.84 ^{jA}	3.65 ^{hB}
Kampferol	2.21 ^{jB}	9.52 ^{hA}	1.90 ^{kC}
Hispertin	46.47 ^{eB}	81.49 ^{dA}	27.05 ^{dC}
Apegnin	0.32 ^{lB}	1.94 ^{kA}	0.20 ^{lC}
7-OH flavon	0.83 ^{kC}	1.17 ^{lB}	1.98 ^{iA}

- ND: Not Detected

- Means followed by different small letters in the same column (effect of variety on concentration of flavonoids) are significantly by Duncan's multiple test ($P \leq 0.05$)

- Means followed by different capital letters in the same row of same parameter (effect within variety) are significantly by Duncan's multiple test ($P \leq 0.05$)

Effect of total soluble solids% (Brix) of sorghum syrup on bioactive compounds

Table 6 and 7 revealed that there was a reversible relationships between phenolic content and TSS, with exceptions for certain compounds such as gallic acid that increased as TSS increased from 73 to 78° and 4-aminobenzoic acid, chlorogenic acid, caffeic acid, vanillic acid, ellagic acid, and coumarin, which exhibited increased levels at TSS values of 73° or 75° Brix compared to other concentrations. Among all treatments, pyrogallol was the predominant phenolic compound. The observed increase in phenolic content can be attributed to the inactivation of polyphenol oxidase, an enzyme responsible for polyphenol degradation during heating. Regarding flavonoid content, a positive correlation with TSS was observed up to 75° Brix. However, further increasing TSS to 80° Brix led to a

decline in both phenolic and flavonoid compounds. Apigenin demonstrated the lowest flavonoid content across all treatments, while Catechin exhibited the highest levels at (75°, 78° and 80° brix) meanwhile, Naringin exhibited the highest level among flavonoid compounds at 73° brix. These findings align with Palermo et. al. (2014) who stated that variation in phytochemical (increasing or decreasing) after cooking might be due to two opposite effect first: thermal degradation that reduce their concentration, and the softening effect of matrix that make phytochemical extractability easier so a higher concentration is observed. Generally, the effect of cooking on phytochemical concentration depends on many factors such as processing considerations (method and time, the structure of food matrix and finally the chemical nature of the particular compound.

Table 6. Effect of total soluble solids% (TSS%) of sorghum syrup on phenolic acids

TSS%	73	75	78	80
Phenolic compound (ppm)				
Gallic	26.51 ^{kC}	81.17 ^{eB}	90.75 ^{dA}	5.09 ^{oD}
Pyrogallol	1022.87 ^{aA}	565.52 ^{aC}	716.54 ^{aB}	472.92 ^{aD}
4-Amino-benzoic	28.99 ^{iB}	39.66 ^{iA}	22.65 ^{kC}	21.04 ^{iD}
Chlorogenic	59.62 ^{hC}	192.47 ^{cA}	106.79 ^{eB}	53.11 ^{dD}
Rosmarinic	6.93 ^{mB}	7.01 ^{nA}	5.83 ^{nC}	5.51 nD
Caffeine	128.87 ^{dA}	27.96 ^{kB}	27.58 ^{iC}	17.37 ^{iD}
P-OH- benzoic	305.22 ^{bA}	64.14 ^{gC}	75.75 ^{gB}	41.95 ^{eD}
Caffeic	29.08 ^{iC}	42.81 ^{hA}	40.98 ^{iB}	14.94 ^{kD}
Vanillic	36.92 ^{gC}	135.78 ^{cA}	78.51 ^{fb}	32.36 ^{fd}
Ferulic	275.89 ^{cA}	74.37 ^{fb}	42.18 ^{hC}	24.10 ^{hD}
Ellagic	105.87 ^{eC}	143.42 ^{bA}	119.51 ^{bB}	96.55 ^{bD}
Benzoic	63.76 ^{fc}	109.96 ^{dA}	84.97 ^{eB}	58.45 ^{cD}
Salicylic	34.84 ^{hB}	40.54 ^{iA}	12.71 ^{mD}	26.51 ^{gC}
Coumarin	7.00 ^{lD}	18.31 ^{lA}	14.97 ^{lB}	12.25 ^{lC}
3-OH-Tyrosol	ND	14.33 ^{mA}	4.93 ^{oC}	7.17 ^{mB}

- ND: Not Detected

- Means followed by different small letters in the same column (effect of brix on Phenolic compound concentration) are significantly by Duncan's multiple test ($P \leq 0.05$).

- Means followed by different capital letters in the same raw of same parameter (effect within brix) are significantly by Duncan's multiple test ($P \leq 0.05$)

Table 7. Effect of total soluble solids% (TSS%) of sorghum syrup on flavonoids

Flavonoids (ppm)	TSS%	73	75	78	80
Naringin		156.95 ^{aA}	118.04 ^{bB}	113.03 ^{bC}	81.59 ^{bD}
Rutin		92.52 ^{cA}	38.18 ^{fB}	23.95 ^{fC}	16.20 ^{fD}
Hisperdin		44.09 ^{fD}	96.33 ^{cA}	68.25 ^{cB}	57.46 ^{cC}
Catecol		136.09 ^{bA}	21.15 ^{hB}	12.77 ^{hC}	4.46 ^{hD}
Catechen		58.69 ^{dD}	219.90 ^{aA}	203.33 ^{aB}	106.43 ^{aC}
Quercetrin		52.09 ^{eB}	53.65 ^{eA}	40.79 ^{dC}	23.63 ^{dD}
Quercetin		7.80 ^{hD}	54.50 ^{dA}	35.78 ^{eB}	14.42 ^{gC}
Narengenin		6.84 ^{iB}	7.26 ^{iA}	6.25 ^{iC}	2.68 ^{iD}
Kampherol		9.52 ^{gD}	24.82 ^{gA}	15.34 ^{gC}	17.11 ^{eB}
Apegnin		1.49 ^{iA}	1.66 ^{iC}	1.72 ^{jB}	0.67 ^{jD}

- ND: Not Detected

- Means followed by different small letters in the same column (effect of brix on flavonoid compound concentration) are significantly by Duncan's multiple test ($P \leq 0.05$).

- Means followed by different capital letters in the same raw of same parameter (effect within brix) are significantly by Duncan's multiple test ($P \leq 0.05$)

4. Conclusion

The optimal selection of sweet sorghum variety and harvest stage is crucial for maximizing the phytochemical benefits derived from the resulting juice or syrup. Phenolic and flavonoid compounds generally exhibited higher levels at the dough stage compared to the milk stage. The concentration process significantly enhanced phenolic content, particularly in the Rex variety, while flavonoids also increased up to a TSS of 75°. Sweet sorghum syrup is rich in polyphenol compounds (pyrogallol, Ellagic, chlorogenic, cateche and naringin). These findings underscore the importance of careful consideration of variety, harvest timing, and processing parameters to produce sweet sorghum products with optimal nutritional value. This research indicates that sorghum cultivated in temperate climates could be a valuable source of bioactive compounds, adding to its nutritional value and potential health benefits. Finally, we recommended that the constant supply/consumption of sweet sorghum syrup as a traditional recipes or as a food ingredients can play an important role in the prevention/ reduction of some chronic diseases i.e. cancer, diabetes, cardiovascular diseases, and inflammatory processes due to presence of phenolic compounds (flavonoids, phenolic acids) provide biochemical and molecular mechanisms to reduce free radicals induced by oxidative stress.

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